

An Investigation by LA-ICP-MS of Possum Tooth Enamel as a Model for Identifying Childhood Geographical Locations of Historical and Archaeological Human Remains from New Zealand

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ABSTRACT

LA-ICP-MS (laser ablation-inductively coupled plasma-mass spectrometry) has been used to analyse enamel from the teeth of brushtail possum (*Trichosurus vulpecula*) in order to model a method for identifying the childhood geographical origin of human remains within New Zealand. The model application of the method is promising for establishing locations of historical and archaeological human remains, including preserved heads, upoko tuhi.

Keywords: LA-ICP-MS, tooth enamel, brushtail possum, geographical origin, archaeological remains, New Zealand

INTRODUCTION

Tooth enamel is a composite mineral with a crystalline structure and it is the most mineralised of the three layers of which teeth are composed: dentine, cementum and enamel. Enamel is 96–97% inorganic, and consists predominately of hydroxyapatite, $\text{Ca}_5(\text{PO}_4)_3\text{OH}$ crystals. The ~3–4% of enamel that is not inorganic consists generally of water and traces of organic matter such as proteins and lipids. Calcium and phosphorus are the main elements found in tooth enamel and these elements can be substituted by a range of trace elements, which are then incorporated into the enamel (at either the cationic centre (Ca^{2+}) or the anionic centres (PO_4^{3-} or OH^-)) at the time of exposure. Major cations that are usually incorporated are Na^+ , K^+ and Mg^{2+} , whereas the common anions are CO_3^{2-} , F^- and Cl^- . However 40 more ions are reported to have been found in the 1000 ppm region, including zinc, iron, aluminium and strontium, and more again in the

100 ppb region, including nickel, lithium, silver, and arsenic (Kang *et al.* 2004). In humans, formation of enamel in the deciduous incisors starts three months after conception and ends one month after birth, and thus can reflect changes *in utero* (Uryu *et al.* 2003); maturation of enamel in other deciduous teeth continues after birth and reflects dietary changes such as weaning (Humphrey *et al.* 2008). After the deciduous teeth fall out the mature, adult teeth, that replace them, have already-formed enamel before they emerge from the gum. Thus the enamel of adult teeth, which does not undergo significant change once matured, represents the environmental exposure between the first month after birth and the time when the adult tooth emerges (Smith 1998). Other mammals can be expected to have analogous developmental phases for enamel.

Inductively coupled plasma-mass spectrometry (ICP-MS) is an established technique for the elemental analysis of both teeth and bones from archaeological samples. A method, commonly described in the literature, to introduce analyte to the instrument is as a solution which is obtained by digesting the sample in a mineral acid. This method was used to estimate lead, zinc and strontium levels in permanent teeth from a Bronze Age tell in the United Arab Emirates, and from an 18th century African burial ground in New York City, and the results were compared with values on deciduous teeth from modern Egypt (Webb *et al.* 2005). Solution ICP-MS has been used in conjunction with isotope analysis to determine the immigrant status of individuals from a grave site in Vanuatu (Bentley *et al.* 2007), and to attempt to determine migration

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patterns of the Lapita people of 3300–2200 BP (Shaw *et al.* 2009). Solution ICP-MS has also been used in a number of modern studies to assess the incorporation into teeth of environmental contaminants such as lead (see for example Arruda-Neto *et al.* 2009; Robbins *et al.* 2010).

The use of laser ablation for sample introduction to ICP-MS (LA-ICP-MS) confers a number of advantages over the use of solution sample introduction. These include the substantially less destructive nature of the technique because of the extremely small sample size required, the ability to acquire chronological data by line scanning, the reduced sample preparation time through the absence of a requirement to separate enamel from underlying dentine and the rapidity of the technique (Lochner *et al.* 1999; Galiová *et al.* 2010). A study of strontium ratios in modern rodent teeth showed no significant difference in results between solution and LA-ICP-MS methods (Copeland *et al.* 2008). A study of lead/calcium ratios in modern teeth also discovered no significant difference between the two techniques (Uryu *et al.* 2003).

LA-ICP-MS has been used to study strontium ratios in the enamel of a prehistoric tooth and by comparison with bones and dentine to determine migration (Prohaska *et al.* 2002). The technique has also been used to determine migration patterns of a prehistoric bear (Galiová *et al.* 2010); to measure the lead content of teeth from an archaeological burial site in New York (Bellis *et al.* 2009; Bellis *et al.* 2008); to determine the feasibility of distinguishing individuals for forensic purposes or from mass burial sites using teeth and bones (Castro *et al.* 2010); to attempt to identify individual remains as members of the Mozart family (Stadlbauer *et al.* 2007); to identify one individual as an immigrant to a community in 5–6th century AD Austria (Prohaska *et al.* 2002); and to determine whether significant diagenesis occurred in the enamel of ancient tooth samples by comparing modern teeth with samples from British Iron age burial sites (Budd *et al.* 1998). Application of LA-ICP-MS to modern materials has been used in a study to examine pre- and post-natal uptake of lead into deciduous teeth in human children (Arora *et al.* 2006). Exposure of infant humans to lead *in utero* was studied by analysing the enamel of deciduous incisors by LA-ICP-MS (Uryu *et al.* 2003). Variations in strontium/calcium ratios of deciduous canines or second molars have been correlated with weaning of infants (Humphrey *et al.* 2008), and this method was intended for exploring weaning practices represented in archaeological samples. Variations of the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio over a whole tooth length were found to reflect the mobility of herbivores in Kruger National Park for over a year indicating the possibilities of this technique for reconstructing historic migrations (Balter *et al.* 2008).

Our particular interest is in establishing the childhood geographical locations of Maori human remains that have been retained as historical or archaeological items or samples, notably preserved human heads (upoko tuhi). A trade between Māori and Europeans in preserved heads was es-

tablished as early as 1770 and continued until the 1830s. Pomare and Hongi Hika of Ngāpuhi, Bay of Islands, were two amongst a number of war leaders who obtained many of the upoko tuhi that were traded. Some were taken from captured settlements and can be traced by moko pattern to their homelands, but others were obtained by capturing prisoners and tattooing them before decapitation. As the moko patterns were those of the captors, the birthplaces of the individuals cannot be ascertained. Ngāpuhi and their allies focussed their attention on the North Island, raiding as far southeast as Hawke's Bay. Fewer Ngāpuhi raids with the death of Pomare in 1826 and of Hongi Hika in 1828, and a proclamation by the governor of the colony of New South Wales in 1831, which banned the traffic in heads, brought about a decline of the trade (Paterson 2010; Te Awēkotuku 2004; Te Awēkotuku *et al.* 2007).

Upoko tuhi were usually taken back to Europe to be placed in museums or in private collections. Since 1992 there have been efforts to recover upoko tuhi and other ancestral remains and since 2003 the Museum of New Zealand Te Papa Tongarewa has acted as the government agent for these returns. There have been ~360 returns including heads from 13 foreign countries and 55 different institutions since 1987. Eighty five upoko tuhi are presently held in Te Papa Tongarewa and possibly an equal number are still held overseas as repatriation efforts continue. (Patterson 2010). The means by which the regions from which the upoko tuhi originated, and to which they will subsequently be returned, is the subject of debate. A non-destructive method that can identify the childhood origin of the individuals from whom the upoko tuhi originated would thus be a valuable asset.

Brushtail possums were chosen as model animals for this study as they are an official pest species distributed throughout New Zealand and are thus trapped in large numbers. More importantly, possum browse local vegetation and, outside urban areas, do not have access to imported foodstuffs or supplements such as might be consumed by humans or domestic animals. The home range of a possum varies between 1.3–1.9 ha, and on average a juvenile possum only moves 5 km from its place of birth, although exceptional females have been measured moving 32 and 41 km (Hutchings 2009). As a result of this, the area in which a possum is ultimately trapped should represent the area and thus the food the juvenile possum ate when the tooth enamel was being formed.

EXPERIMENTAL METHODS

Samples

Possum jaws were obtained from the following regions: Auckland, Christchurch, Coromandel, Nelson, Okuru (S Westland), Taumarunui, Taupo, the West Coast of the South Island and Whangarei. Locations are taken from the New Zealand Map Grid using projected coordinates based

upon the New Zealand Geodetic Datum 1949 (NZGD49) (for conversion to other systems see <http://apps.linz.govt.nz/coordinate-conversion/>) and information on soils was obtained from the regional land use section of Te Ara, the New Zealand Government online Encyclopaedia of New Zealand (<http://www.teara.govt.nz/>).

For the Auckland region four jaws were obtained from Waiuku (E 2660374, N 6433557) (E 2660579, N 6434542) (E 2660822, N 6433810) (E 2660374, N 6433557). Four more jaws were sourced from Awhitu (Andrew Pye Road), a small community about 40 km from Waiuku (E 2657239, N 6458324). Both of these sets of samples come from coastal sedimentary soils. Due to proximity to coastlines, these soils differ greatly from the typical soils found in the greater Auckland region and are made up of basalt, sandstone and marine pumice. Iron sands on the western side of Waiuku could also contribute minerals such as Fe and Mn to soils.

Five jaws were obtained from the Christchurch region (E 2414975, N 5763475) (E 2434025, N 5755550) (E 2470650, N 5748275) (E 2476550, N 5768975) (E 2473125, N 5771300). This region has greywacke sedimentary soils strongly influenced by nearby rivers and seas.

Five samples were obtained from the Coromandel region; three of the five were sourced from Waitete Bay (E 2730200, N 6501678) whereas the other two were from Papaaroa (E 2733000, N 6498150). The Coromandel soil type derives from the Coromandel volcanic zone and marine sediment rock is also present. The specific area fits into the Te Mata subgroup which contains imbedded sandstone and mudstone and a variety of soil types.

Three jaws were obtained from the Nelson Department of Conservation (E 2505820, N 5991690) (E 2580320, N 5990160) (E 2580320, N 5990160). The samples were taken from an area close to Blenheim, where the soil types are mostly made up of volcanic basalt rocks, specifically Mandamus Igneous Complexes. There may also be present sandstone and mudstone soils, as well as some soils commonly found near coastal areas. Soils in this region are generally deficient in copper, cobalt, nitrogen, phosphorus, boron, and magnesium.

Six jaws were obtained from Taumarunui (E 2697724, N 6258592) (E 2697697, N 6259392) (E 2695908, N 6257754) (E 2696279, N 6253599) (E 2702951, N 6255754) (E 2695797, N 6255479); the samples were taken from an area where the soils are predominately from the Taumarunui group formation. These soils are thin to medium bedded, and are made up of graded sandstone, mudstone, volcanic basalt and local imbedded limestone.

Six samples were received from the South East side of Lake Taupo (E 2762156, N 6250164). Volcanic pumice showers from the Taupo and Kaharoa eruptions, which occurred in the last 4000 years, form the basis of these soils in which cobalt is deficient as also may be potassium, boron, magnesium, cobalt, copper, sulphur and phosphate.

Six samples were sourced from the West Coast of the South Island, four of them from Otira, west of Hokitika (E 2392663, N 5818671), and two from Whitcombe, north of Hokitika (E 2362312, N 5836626). The area from which these samples were taken is strongly influenced by the numerous rivers around this area, and thus tend to be river gravel and sand soils also known as greywacke soils.

Five samples were obtained from Okuru, north of Fiordland National Park (E 2181593, N 5692028); Okuru soils tend to be greywacke also, rather than the granite type of soils found further south in this region.

Four samples were sourced from Whangarei (Mangapai Caves Road) in farmland outside the central area (E 2628690, N 6590879). There are two possible soil types in this specific area: Waipara group soils, which are predominately made up of volcanic sandstone, basalt and argillite soils; and the Waitemata group soils, which are made up of Mangakahia complex mudstones; the area is also close to the sea so may show marine influence.

Preparation of samples

Jaws were cut away from deceased possums, and flesh was removed using an autoclave. Jaws were placed in beakers with distilled water (200 mL) covered with tin foil and boiled (300°C, 2 h). Flesh and teeth were removed and the teeth were cleaned of any surface impurities (acetone, distilled water) and left to dry. Entire teeth were mounted on glass microscope slides using Blu-Tack® (3 × 5 rows); entire teeth were also used by Copeland *et al.* (2008) in their study of rodent teeth. Three molar teeth from each possum were analysed. The calibration standard was mounted on the same slide.

Instrumentation

LA-ICP-MS was performed on a New Wave UP-213 laser ablation system fitted with a Nd:YAG 213 nm laser (New Wave Research, Fremont, California, USA) and was controlled by New Wave Research – Laser Ablation software. The laser was connected to a PerkinElmer SCIEX ELAN DRC II inductively coupled plasma (ICP) mass spectrometer with a quadrupole mass spectrometer (PerkinElmerSciex, Concord, Ontario, Canada). The ICP mass spectrometer was controlled by ELAN software, version 3.3. Laser and ICP parameters are shown in Table 1. Data was imported into time resolved analysis software, Gemoc Laser ICP-MS Total Trace Elemental Reduction (GLITTER) (version 4.4.1, Macquarie Limited, Australia) for analysis. The data was exported from GLITTER in the Comma Separated Values (csv) format. Calcium was used as an internal standard and the counts in each analysis were standardised to the stoichiometric abundance of CaO in hydroxyapatite; assuming that enamel is 96% hydroxyapatite this gave the value 53.587%.

Table 1. Parameters for operation of the LA-ICP-MS

Laser power	40%
Spot size	60 μm
Repetition rate	10 Hz
Ablation time	60 s
Carrier gas (Helium)	1.00 L min ⁻¹
Nebuliser gas (Argon)	0.65 L min ⁻¹
Plasma gas	15 mL min ⁻¹
Auxiliary gas	1.2 mL min ⁻¹
Laser wavelength	213 nm
RF power	1350 W
Scan mode	Peak hopping
Ablation mode	Single spot
Readings/Replicate	600
Replicates	1

Analysis of samples

The sample slide was placed in the sample chamber and the chamber was purged with argon and then with helium (5 minutes each) to remove oxygen. The laser was fired with the shutter closed at 60% power with a spot size of 60 μm and with a repetition rate of 10 Hz for 30 to 60 minutes until constant power readings were obtained. Instrument drift was corrected by measuring the calibration standard twice at the beginning of a run and twice at the end of the run. Four spots on each tooth were ablated; 40% laser power gave a good signal without breaching the enamel and passing through to the dentine. The following isotopes were analysed sodium (²³Na), magnesium (²⁴Mg), phosphorus (³¹P), calcium (⁴³Ca), manganese (⁵⁵Mn), zinc (⁶⁴Zn), strontium (⁸⁸Sr), barium (¹³⁸Ba). An attempt to reproduce the matrix-matched reference material synthesised in-house by Bellotto & Miekeley (2000) produced an unsatisfactory material. All the data in this study was collected using the standard reference material (SRM) NIST 612 which was purchased from the National Institute of Standards and Technology (USA); a higher laser power of

60% was required to give a good signal from this standard. NIST SRM 612 was used by Kang *et al.* (2004) and by Humphrey *et al.* (2008) and Castro *et al.* (2010) also used it as a calibrant; another glass standard NIST SRM 610 was used by Arora *et al.* (2006).

Statistical analyses

Although the Glitter program produces putative ppm concentrations by standardising isotopes against Ca, we have avoided assuming all samples have the same Ca concentration by calculating the relative proportions of all eight isotopes including calcium. The resulting data is thus compositional and needs special treatment as explained in Aitchison (1986) and illustrated in Campbell *et al.* (2009). In particular any treatment of the variability in the data, or of the interrelationships between the elements should be expressed in terms of a logarithmic transformation of the ratios of variables if standard methods of analysis are to be valid. In particular for principal component analysis we have used centred log ratios and, for discriminant analysis and the calculation of canonical variates, additive log ratios (Campbell *et al.* 2009). Multivariate analyses and the production of descriptive displays were carried out using Minitab (2010) and GenStat (2009) software.

RESULTS AND DISCUSSION

Using the raw output from Glitter (ppm), any reading below the minimum detection limit (MDL) was assigned a value of MDL/2 (Campbell *et al.* 2009). Table 2 illustrates the average eight-element compositions observed in the nine regions; the compositional data per region is summarised by taking arithmetic means of the proportions across all teeth within each region; Campbell *et al.* (2009) suggest using geometric means but in fact there is little difference in the results. As expected, calcium (⁴³Ca) and phosphorus (³¹P) are the main elements present and sodium (²³Na) is the principal trace element. The ratio of calcium to phosphorus of ~1.47 does not match that expected for tooth

Table 2. Proportions of the individual elements by regions; the data for each region is constrained to sum to 1

	⁴³ Ca	²³ Na	²⁴ Mg	³¹ P	⁵⁵ Mn	⁶⁴ Zn	⁸⁸ Sr	¹³⁸ Ba
Auckland	0.6512	0.016969	0.001201	0.3287	0.0003704	0.0003799	0.0006472	0.0001409
Christchurch	0.5380	0.008087	0.001917	0.4504	0.0004362	0.0005189	0.0004568	0.0000552
Coromandel	0.5886	0.007494	0.003459	0.3973	0.0015501	0.0004793	0.0008261	0.0001903
Nelson	0.4335	0.006241	0.002136	0.5541	0.0002375	0.0003557	0.0006681	0.0001457
Okuru	0.6071	0.007702	0.003672	0.3798	0.0001600	0.0005165	0.0005027	0.0003160
Taumarunui	0.6117	0.006893	0.002711	0.3767	0.0001172	0.0007714	0.0007778	0.0001740
Taupo	0.5187	0.008273	0.003472	0.4678	0.0001005	0.0005317	0.0006878	0.0003458
West Coast	0.5293	0.009363	0.003337	0.4563	0.0002353	0.0006658	0.0004940	0.0001219
Whangarei	0.6009	0.010294	0.001631	0.3851	0.0004509	0.0009615	0.0004891	0.0001509
All	0.5748	0.009585	0.002599	0.4110	0.0003951	0.0005725	0.0006229	0.0001831

enamel, based on the formula for hydroxyapatite, of ~2.15. Although several of the works cited herein normalise to calcium none actually give values for both calcium and phosphorus, which would permit comparison. Lower than expected ratios of calcium to phosphorus in surface enamel are a phenomenon that has been observed for a long time and with different techniques. Weatherell (1975) attributed it to calcium deficient hydroxyapatite structures. Lou *et al.* (2009) in a study of enamel surface relative to dental practice found a mean calcium to phosphorus ratio of 1.41 using X-ray photo-electron spectroscopy, a surface technique.

Individual value plots (every spot on each tooth) of each element by region were created to allow simple visual analysis (Campbell *et al.* 2009), Figure 1 shows, as an ex-

ample, every spot for Mg using the log ratio form of means for the Taumarunui samples.

Subsequent analyses were based on geometric mean proportions across spots for each jaw. This gave compositional data for 143 teeth from 49 jaws. A compositional variation array (Aitchison 1989) is given in Table 3; the top right hand triangle of the table was prepared by taking the mean logarithmic ratios, for example mean ($\ln\{Ca/Ba\}$) which indicates relative proportions. The bottom left hand triangle contains the standard deviation of the log ratios, for example $SD(\ln\{Ca/Ba\})$, and is thus an expression of where variation in the composition proportions originates. Figure 2 illustrates some features of the first row and first column of Table 3; note that Mn (with mean $\ln\{Ca/Mn\} = 8.4$) and Sr (mean log ratio 6.9) are present at similar

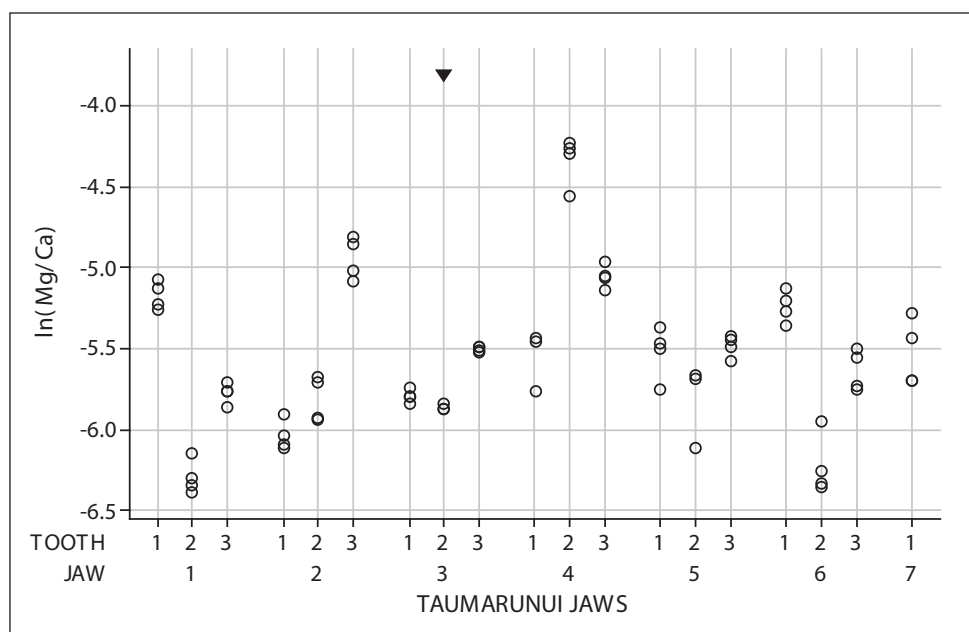


Figure 1. Individual value plots for magnesium ($\ln\{Mg/Ca\}$), for Taumarunui; a potential outlier is marked

Table 3. Variation matrix of elements in the dataset, the upper right triangle shows the mean loge ratios of each two element combination and the lower left triangle indicates the standard deviation of the loge ratios

	⁴³ Ca	²³ Na	²⁴ Mg	³¹ P	⁵⁵ Mn	⁶⁴ Zn	⁸⁸ Sr	¹³⁸ Ba
⁴³ Ca		4.17	5.66	0.34	8.39	7.05	6.94	8.45
²³ Na	0.35		1.49	-3.82	4.23	2.88	2.77	4.29
²⁴ Mg	0.68	0.77		-5.31	2.74	1.39	1.28	2.79
³¹ P	0.29	0.47	0.65		8.05	6.71	6.59	8.1
⁵⁵ Mn	1.52	1.61	1.80	1.59		-1.35	-1.46	0.06
⁶⁴ Zn	0.61	0.79	1.00	0.59	1.64		-0.11	1.40
⁸⁸ Sr	0.47	0.56	0.61	0.53	1.68	0.87		1.51
¹³⁸ Ba	0.82	0.86	0.73	0.87	1.85	1.12	0.62	

levels but at much lower levels than that of P (mean log ratio 0.34). However there is much more variability in the ratio of Mn to Ca across the data set ($SD = 1.52$) than in the ratios of P and Sr to Ca ($SD = 0.29$ and 0.47 , respectively).

Principal component analysis (PCA) was used to condense the description of variation in the data; the first principal component explained 32% of the variation across all teeth. However Table 4 shows that the majority of variation in that component arises between teeth in the same jaw. Most of the literature describes analyses of single teeth from individuals or does not specifically compare data from different teeth types in the same individual; Arora *et al.* (2006) have indicated that this is an area that requires further examination. Second and third molars presumably from the same individual impala showed no significant differences in strontium/calcium ratios or in strontium

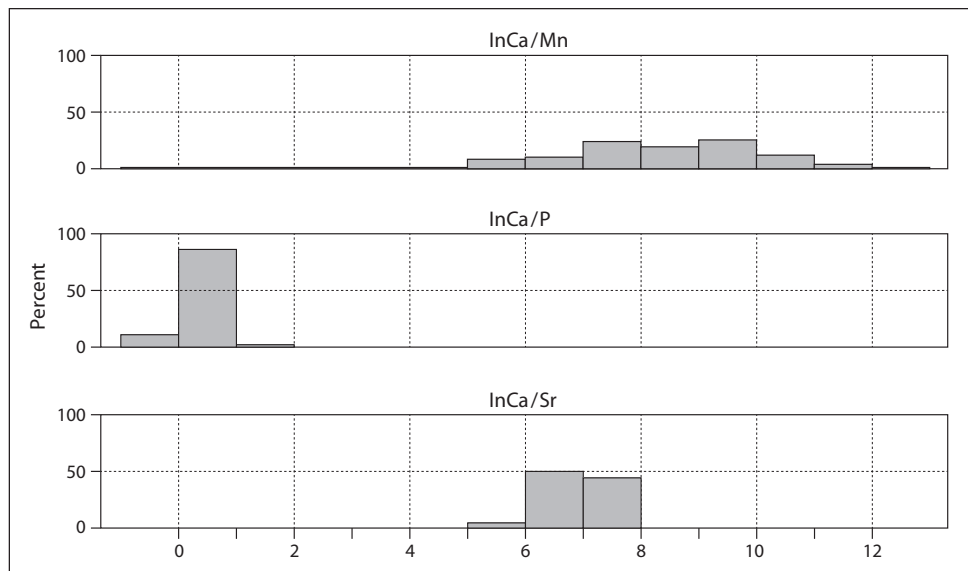


Figure 2. Some features of the compositional variation array; note that strontium (Sr) and manganese (Mn) are present in similar amounts but at a lower percentage than phosphorus (P); however manganese shows a greater variability than either strontium or phosphorus.

Table 4. Comparison of origin of variation in PCA

	Score 1/PC1			Score 2/PC2		
	Variance components	% of total	SD	Variance components	% of total	SD
Location	0.328	12.81	0.573	0.412	16.18	0.642
Jaw	0.396	15.45	0.629	0.705	27.68	0.840
Tooth	1.838	71.74	1.356	1.431	56.14	1.196

isotope ratios (Balter *et al.* 2008). However Humphrey *et al.* (2008) examined two teeth (canine and molar) from each of two individuals as part of a study looking at rates of change of normalised strontium intensities and, although chronological trends matched, absolute rates between the different teeth types varied. Castro *et al.* (2010) examined two mandibular third molars and a maxillary third molar from the same individual looking at enamel alone and also the entire tooth. PCA of the latter results showed closeness between the two mandibular molars but significant separation between these and the maxillary molar; this was identified by the authors as one of a number of issues requiring more study. In studies relating to human dentistry both Lou *et al.* (2009) and Weatherell *et al.* (1974) identified significant variation in element distribution in enamel of different teeth and from different locations on the same tooth and the former recommends examination of the same tooth and the same location within the tooth in order to facilitate comparison between individuals.

In the entire data set, 86% of the variation was explained by the first four principal components. The third component was particularly effective at separating the

Auckland region (Fig. 3).

Linear discriminant analysis (LDA) using additive log ratios was used to attempt to find functions of the elemental proportions which organise the data into groups (geographical origins). Table 5 shows the cross-validated LDA results for all regions with an overall proportion of correctly identified samples of 0.748; that is 107 samples from 143 were correctly placed in their region of origin. The results are illustrated in a plot of the canonical variates (Fig. 4), which underlie the analysis, and show that the Nelson region is completely separated from all other regions and that the Auckland region is largely separated from other regions. The first canonical variate is highly correlated with the log ratio of phosphorus (P) and sodium (Na), as is the third principal component.

When considering application of this methodology to for example upoko tuhi, it is possible to take into account historical evidence. The major regions that overlap with others are Okuru, Christchurch and the West Coast and so it was felt that, based upon the activities of the principal parties involved in the trade of upoko tuhi (Te Awēkotuku 2004; Te Awēkotuku *et al.* 2007), it would be reasonable to remove these three regions in order to simplify the analysis. Table 6 shows the cross-validated LDA results for the regions excluding Christchurch, Okuru and the West Coast and the plot of canonical variates is illustrated in Figure 5. The overall proportion of samples correctly identified was 0.885 that is 85 correct identifications from 96 samples and the plot shows markedly improved separation.

The samples designated as Auckland actually fall upon the boundary between Tai Tokerau and Tainui regional groupings and the former regional grouping would en-

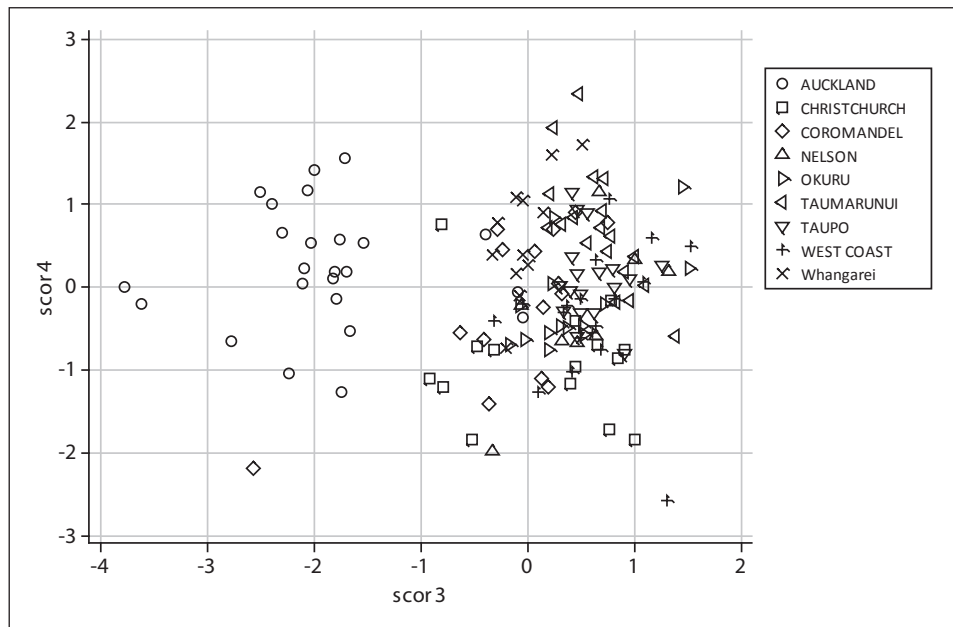


Figure 3. Scatterplot of the 3rd and 4th principal components showing good separation for the Auckland region afforded by the 3rd component

Table 5. Cross-validated LDA of all regions showing the proportion of samples correctly assigned

	Auckland	Christchurch	Coromandel	Nelson	Okuru	Taumarunui	Taupo	West Coast	Whangarei
Auckland	21	0	0	0	0	0	0	0	0
Christchurch	0	9	1	0	1	0	1	2	0
Coromandel	0	3	10	1	0	3	0	0	0
Nelson	0	1	0	7	0	0	0	0	0
Okuru	1	0	0	0	12	2	0	1	0
Taumarunui	0	0	2	0	1	13	0	0	0
Taupo	0	0	1	0	0	0	13	3	0
West Coast	0	2	0	0	0	0	4	10	0
Whangarei	2	0	1	0	0	1	0	2	12
Total N	24	15	15	8	14	19	18	18	12
N correct	21	9	10	7	12	13	13	10	12
Proportion correct	0.875	0.600	0.667	0.857	0.857	0.684	0.722	0.556	1.000

compass the Whangarei samples as well. Nevertheless the sampling regions are geologically separated so the minor overlap that occurs would probably be reduced even more by expanding the sample sizes in both regions. The Taupo and Taumarunui samples, which also show overlap, fall into the Arawa and Whanganui regional groupings respectively but once again the minor overlap might be reduced further by expanding the sample base.

When considering human remains it would not be necessary or even practical to examine entire teeth. Possum teeth are so small that the most practical way to sample is to use entire teeth, as did Copeland *et al.* (2008) with rodent teeth. With larger teeth, small sections of enamel

can be separated from the tooth and analysed. The laser ablation technique requires only a tiny amount of material and with precious objects, such as upoko tuhi, sections of enamel could be removed by an individual skilled in dentistry to minimise the impact of sampling.

We conclude from this preliminary study that this methodology shows considerable potential for use in identifying geographical origins of human remains within New Zealand either alone or as an adjunct to other techniques. However, a greatly expanded database, both geographically and numerically, is required to clarify points of overlap and to ensure that traditional regional groupings are adequately represented.

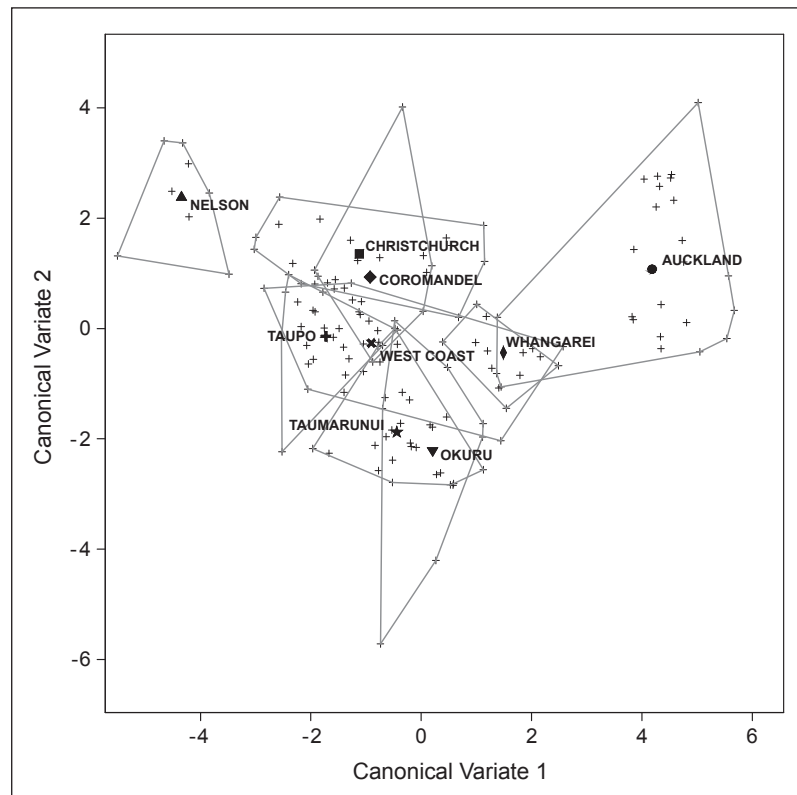


Figure 4. Plot of canonical variates for all regions

Table 6. Cross-validated LDA of selected regions showing the proportion of samples correctly assigned

	Auckland	Coromandel	Nelson	Taumarunui	Taupo	Whangarei
Auckland	21	0	0	0	0	0
Coromandel	0	12	1	2	1	0
Nelson	0	0	7	0	0	0
Taumarunui	0	2	0	16	0	0
Taupo	0	1	0	0	17	0
Whangarei	3	0	0	1	0	12
Total N	24	15	8	19	18	12
N correct	21	12	7	16	17	12
Proportion correct	0.875	0.800	0.875	0.842	0.944	1.000

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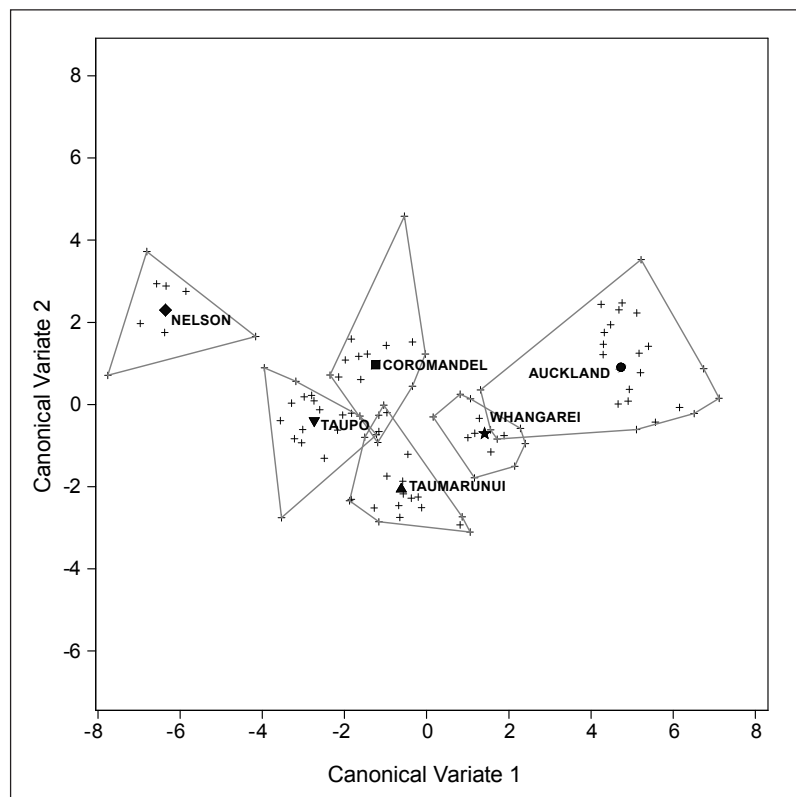


Figure 5. Plot of canonical variates for selected regions.

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